*Bulgarian Journal of Veterinary Medicine* (2004), 7, No 1, 57–62

# SPECIFIC OPTICAL ACTIVITY AND 5-HYDROXYMETHYL-2-FURALDEHYDE CONTENT IN HONEY FROM BEE COLONIES FED UP WITH SUGAR SOLUTION AND ISOSWEET 77555 P

D. DINKOV[1], I. JELYAZKOVA[2], V. RUSSEV[1]& I. VACHIN[1]

[1]Faculty of Veterinary Medicine, [2]Faculty of Agriculture, Trakia University;
Stara Zagora, Bulgaria

**Summary**

Dinkov, D., I. Jelyazkova, V. Russev and I. Vachin, 2004. Specific optical activity and 5-hydroxymethyl-2-furaldehyde content in honey from bee colonies fed up with sugar solution and Isosweet 77555 P. *Bulg. J. Vet. Med.*, **7**, No 1, 57–62.

Bee colonies, equalized by the method of analogues, reared freely and in isolated apiaries, were additionally fed up stimulating or intensive with sugar solution and Isosweet 77555P (IST). It was found out that the specific optical rotation (SOR) of honey increased following the stimulating and especially the intensive feeding up of bee colonies. Honey SOR was higher in bee colonies fed up with IST compared to those with sugar solution. The 5-hydroxymethyl-2-furaldehyde (hydroxymethylfurfural, HMF) content of honey after the stimulating and intensive feeding up of bee colonies with IST and sugar solution did not exceed the maximum allowed quantity for unprocessed honey. The honey HMF content decreased after the intensive feeding up of bee colonies with both IST and sugar solution regardless of the rearing method. In isolated reared bee colonies, fed up only with IST, honey HMF was found out to increase after the stimulating feeding up.

**Key words**: bee feeding up, bee honey, Isosweet 77555 P, 5-hydroxymethyl-2-furaldehyde (HMF), specific optical rotation (SOR), sugar solution (1:1)

## INTRODUCTION

The specific optical rotation of nectar honey is generally negative and very rarely positive (Chudakov, 1963; Sacchi and Bosi, 1963; Battaglini and Bosi, 1973; Persano et al., 1995). All types of nectar honey in Bulgaria have a negative SOR (Ivanov and Mitev, 1972; Ivanov, 1978; 1986) whereas the honey from bee colonies, intensively fed up with sugar solution (the so-called "sugar honey") and the honeydew honey – positive SOR. When glucose, dextranes or starch are added to the nectar honey, SOR is found out to change from negative to positive (Ivanov, 1986).

The "sugar honey" has a lower, while honey, heated continuously at a high temperature or adulterated with glucose or inverted sugar – higher HMF content (Ivanov, 1978; 1986), The changes in HMF levels were used as indication of adulteration of honey with IST (Dinkov, 2001).

After the feeding up of bees with sugar solution and IST, deviations in the organoleptic and most physicochemical indices of honey are observed (Dinkov *et al.*,

2002; Jeliazkova *et al.,* 2002; Dinkov *et al.,* 2003). According to some investigators (Ivanov, 1978; White, 1980; Wehling *et al.*, 1997) the feeding up of bees with sugar solutions influence both the SOR and HMF content of honey. Therefore, the adulterations of honeys and the feeding up of bees with sugar altered honey SOR and HMF concentrations, but there are no comparative data showing their changes when bees are fed up with sugar solution or IST.

The aim of the present study was to observed the effect of feeding up of bees with sugar solution or IST upon the specific optical rotation and the HMF content in bee honey.

## MATERIALS AND METHODS

Eight bee colonies, placed in Dadan-Blat hives, equalized by the method of analogues (age of queens, bees from 2.5 to 2.75 kg and from 52000 to 54500 cells with capped worker brood) divided into 4 groups (2 colonies each) were used. Three groups of bee colonies were reared freely under natural conditions and another one – in 2×2×2 m cells with access to water in outer water troughs.

The feeding up of bee colonies was performed with sugar solution (1:1) and Isosweet 77555 P (IST) – a purified and concentrated aqueous solution of saccharides (55% fructose and 40% glucose), produced by enzyme hydrolysis of maize starch in "Amilium – Bulgaria" Ltd., Razgrad. The lower water content of IST (23.60%) necessitated its dilution with water to 25.65%, equal to that of sugar solution.

The experiment was performed in a private apiary in August 2001 following the design presented in Table 1. At the end of the stimulating (August 9) and the intensive (August 19) feeding up, 5 combs were removed from each bee colonies. From each comb, honey samples of about 25–30 g were obtained by draining some previously decapped parts. Thus, 10 samples were obtained from each group.

The analyses were carried out in the laboratory of the Department of Hygiene, Technology and Control of Animal Foodstuffs, Veterinary Legislation and Man-

**Table 1.** Experimental design

| Conditions | Groups of bee colonies | | | |
|---|---|---|---|---|
| | I (controls) | II | III | IV |
| Number of bee colonies | 2 | 2 | 2 | 2 |
| Method of rearing | Free | Free | Free | Isolated |
| Stimulating feeding between 31.07. – 09.08.2001 | – | Sugar solution 200 mL /24 h per colony | IST 200 mL/24 h per colony | IST 200 mL/24 h per colony |
| Intensive feeding between 10.08. – 19.08.2001 | – | Sugar solution 500 mL /24 h per colony | IST 500 mL/24 h per colony | IST 500 mL/24 h per colony |

agement, Faculty of Veterinary Medicine, Trakia University, Stara Zagora. The SOR and the HMF content were assessed according to the methods determined by the European Honey Commission (Bogdanov *et al.,* 1997).

The statistical analysis between groups was performed by the Student's t-test.

When SOR data ($[\alpha]_D^{20}$) were interpreted, the term "increase" meant values closer to zero (for negative values) and the term "decrease" – values closer to zero (for positive values).

## RESULTS AND DISCUSSION

The results of honey analyses are presented in Table 2. They show that within the period of active honey collection (August 10–19, 2001) the SOR of honey obtained from the control group decreased slightly from –20 to –22 $[\alpha]_D^{20}$.

After the stimulating and intensive feeding up with sugar solution, the SOR of honey from group II was higher vs control honey, more pronounced after intensive feeding up (–13 and –10 $[\alpha]_D^{20}$ vs –20 and – 22 $[\alpha]_D^{20}$respectively) ($p<0.05$ after intensive feeding up).

The SOR of honey from group III showed analogous results but with higher mean values of $[\alpha]_D^{20}$ (–5). Following the intensive feeding up the difference vs group I was statistically significant at $p<0.05$.

The honey SOR of group IV after the stimulating (–9 $[\alpha]_D^{20}$) and the intensive (–5 $[\alpha]_D^{20}$) feeding up with IST was statistically higher vs controls. In comparison with group III, only the mean values after stimulating feeding up were different (relatively higher). When compared to group II, the SOR values of group IV were higher both after stimulating and intensive feeding up – twice higher in the latter case.

**Table 2.** Specific optical rotation (SOR) and 5-hydroxymethyl-2-furaldehyde (HMF) content of honey in samples from bee colonies, fed up stimulatingly and intensively with sugar solution (1:1) and Isosweet 77555P. Data are presented as means (SD), n=10

| **Parameters** | Groups of bee colonies | | | |
|---|---|---|---|---|
| | I (control) | II | III | IV |
| *After stimulating feeding up* | | | | |
| Specific optical rotation ($[\alpha]_D^{20}$) | –20.00 (0.11) | –13.00 (13.29) | –13.00 (11.94) | –9.00 (0.11)• |
| HMF content (mg/kg) | 7.29 (6.53) | 11.52 (6.55)• | 5.56 (2.81) | 15.90 (1.09)• |
| *After intensive feeding up* | | | | |
| Specific optical rotation ($[\alpha]_D^{20}$) | –22.00 (0.11) | –10.00 (0.15)• | –5.00 (0.54)• | –5.00 (0.77)• |
| HMF content (mg/kg) | 12.86 (1.11) | 7.48 (6.22)• | 3.84 (1.04)• | 8.06 (0.65)• |

• $p<0.05$ vs control group.

Our average SOR values varied between –5 and –22 $[\alpha]_D^{20}$ and were within ranges (from 0 to –41.0 $[\alpha]_D^{20}$) reported by other authors (Chudakov, 1963; Sacchi and Bosi, 1963; Ivanov and Mitev, 1972; Ivanov, 1978; 1986). The control SOR values (–20 and –22 $[\alpha]_D^{20}$) corresponded to most commonly reported data for Bulgarian nectar honeys (–20.0 and –29.9 $[\alpha]_D^{20}$) (Ivanov and Mitev, 1972; Ivanov, 1978; 1986).

The tendency toward increase in honey SOR in bee colonies fed up with sugar solutions, observed by Chudakov (1963), Sacchi et Bosi (1963), Ivanov and Mitev (1972), Battaglini et Bosi (1973), Ivanov (1978, 1986), Persano *et al.*, (1995) was also present when IST was used. Therefore, it could be assumed that the increased honey SOR could be an evidence for intensive feeding up of bee colonies with IST as well.

The HMF content in control honey for the 10-days-period (from August 10 to 19, 2001) of the active honey collection increased from 7.29 to 12.86 mg/kg. After the stimulating feeding up, honey HMF content in group II (11.52 mg/kg) was significantly higher than that in controls ($p<0.05$), while after the intensive feeding up it was lower (7.48 mg/kg) compared to that in controls ($p<0.05$) and to stimulating feeding up of the same group.

The HMF concentrations in honey from group III decreased slightly after the stimulating (5.56 mg/kg) and considerably ($p<0.05$) after the intensive feeding up (3.84 mg/kg) compared to controls.

In group IV, HMF content was more than doubled after the stimulating feeding up (15.90 mg/kg) than that in controls ($p<0.05$). After the intensive feeding up however, this relation was opposite ($p<0.05$). When both feeding up methods were compared, HMF concentrations were found to be almost twice lower for the intensive (8.06 mg/kg) vs the stimulating feeding up (15.90 mg/kg). The latter value was almost three times higher vs group III (5.56 mg/kg) and slightly higher vs group II (11.52 mg/kg)., and the concentrations after the intensive feeding up – two times higher vs group III (3.84 mg/kg) and similar to group II (7.48 mg/kg).

The average honey HMF content for all bee colonies was within the maximum allowable limits. According to the Bulgarian State Standard 2673 (1989), the acceptable HMF content in unprocessed honey is 20 mg/kg. For processed honey, the European Honey Commission (Bogdanov *et al.* 1997), the German standard (Dustmann, 1994) and the Bulgarian State Standard 2673 (1989) allow up to 40 mg/kg HMF, whereas the German Union of Apiculture – no more than 15 mg/kg HMF (Dustmann, 1994).

According to White *et al.* (1964) and Ivanov (1978, 1986), the HMF levels in unheated natural honey is almost always lower than 10 mg/kg. We observed higher values in the honey after 10 days of active honey collection after stimulating feeding up with sugar solution and after stimulating feeding up with IST in bee colonies reared in isolated apiaries.

The HMF values determined by us were lower after the intensive feeding up with both sugar solution and IST, regardless of the rearing approach. In control group, the opposite tendency was present: HMF content increased for 10 days of active honey collection.

## CONCLUSIONS

The specific optical rotation of honey increased following the stimulating and especially after the intensive feeding up of bee colonies with sugar solution and IST.

Honey SOR was higher in bee colonies fed up with IST compared to those with sugar solution. The honey from colonies reared isolatedly after both stimulating and intensively feeding up with IST showed an increased specific optical rotation. The HMF content of honey after the stimulating and intensive feeding up of bee colonies with IST and sugar solution did not exceed the maximum allowed quantity for unprocessed honey. The honey HMF content decreased after the intensive feeding up of bee colonies with both IST and sugar regardless of the rearing method. In isolatedly reared bee colonies, fed up only with IST, honey HMF was found out to increase after the stimulating and to decrease after the intensive feeding up.

REFERENCES

Battaglini, M. and G. Bosi, 1973. Caratterizzazione chimico-fisica dei mieli monoflora sulla base dello spetro glucidico e del potere rotatio specifico. *Scienza e Tecnologia degli Alimenti*, **3**, 217–221.

Bogdanov, S., P. Martin and C. Lüllman, 1997. Harmonised Methods of the European Honey Commission. *Apidologie*, Extra issue, 1–59.

Bulgarian State Standard 2673, 1989. Bee Honey, Sofia, pp.1–7.

Chudakov, V. G., 1963. Composition and properties of bee honeys in the Soviet Union. In: Proceedings of the XIX International Congress of Apiculture, Moskow.

Dinkov, D, I. Zhelyazkova, I. Vashin and V. Rusev, 2003. Effect of supplementary feeding of bees with sugar solution (1:1) and Isosweet 77555P (IST) on the sensory characteristics of bee honey. *Trakia Journal of Sciences*, **1**, No 1, 22–26.

Dinkov, D., 2001. Experimental detection of honey adulteration with Isosweet 77555 P using a modification of the Winkler's method. *Apiacta*, **36**, No 4, 160–166.

Dinkov, D., I. Jelyazkova, V. Russev and I. Vashin, 2002. Effect of supplementary feeding of bees with Isosweet 77555P on some quantitative characteristics of bee honey. *Animal Sciences* (*Sofia*), **39**, No 4–5, 158–160 (BG).

Dustmann, J. H., 1994. Qualitätsmerkmale und Untersuchungskriterien für Honig im Imker-Honigglas des Deutschen Imkerbundes e.V. *Deutscher Imkerbundes e.V.*, **3.3.**, 2–10.

Ivanov, Ts. and B. Mitev, 1972. Study on the physical and chemical properties and the composition of Bulgarian bee honey, *Agricultural Sciences (Sofia),* **9**, No 8, 111–117 (BG).

Ivanov, Ts., 1978. Studies on the composition and properties of Bulgarian bee honey. PhD thesis, Ministry of Agriculture and Food Industry, Institute of Animal Husbandry – Kostinbrod, Experimental Station of Apiculture – Sofia, pp.153–160.

Ivanov, Ts., 1986. Quality, standartization and description of bee products (a review). Agricultural Academy, Center of Scientific and Technical Information, Sofia, 7–32.

Jeliazkova, I., D. Dinkov, V. Rusev, and I. Vashin, 2002. Effect of feeding bees with sugar solution (1:1) and Isosweet 77555P on some qualitative parameters of honey. In: Proceedings of Scientific Conference with International Participation, June 6–7, vol. 2, Trakia

University Press, Stara Zagora, Bulgaria, 300–303.

Persano, O. L., M. G. Piazza, A. G. Sabatini and M. Accorti, 1995. Characterization of unifloral honeys. *Apidologie*, **26**, 453–465.

Sacchi, R. and G. Bosi, 1963. Variabilita nel contenuto di ceneri, ferro, anidride fosforica, calcio, proteine e della "rotatione specifica" in mielli italiani. *L´Apicolture d´Italia*, **89**, 30.

Wehling, M., J. H. Dustmann und W. von der Ohe, 1997. Konversion definierten Zuckerlösungen nach Aufnahme durch Honigbienen. *Apidologie,* **28**, 164–165.

White, J. W. Jr., 1980. Detection of honey adulteration by carbohydrate analysis. *Journal of Assocociation of Official Analytical Chemists*. **63**, No 1, 11–68.

White J. W. Jr., I. Kushnir and M. H. Subers, 1964. Effect of storage and processing temperatures on honey quality. *Food Technology,* **18**, 153–156.

Paper received 15.05.2003; accepted for publication 04.03.2004

**Correspondence**:

Dr. D. Dinkov
Dept. of Hygiene, Technology and Control of Animal Foodstuffs, Veterinary Legislation and Management,
Faculty of Veterinary Medicine,
Trakia University,
6000 Stara Zagora, Bulgaria